

SHORT COMMUNICATION

Dormancy and germination in three *Acacia* (*Fabaceae*) species from central Argentina

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Abstract

We studied dormancy and germination requirements of seeds of *Acacia aroma*, *A. caven* and *A. furcatispina* from a semi-arid region of central Argentina. Imbibition experiments were performed to determine the rate of water uptake in seeds. To determine optimal temperature for germination, seeds were incubated at three temperature regimes (15/5, 25/15 and 35/20°C) with a 12/12 h daily photoperiod or in total darkness. Additionally, differences in dormancy and germination in seed colour morphs of *A. aroma* were studied. Seeds of *A. aroma* and *A. caven* had impermeable coats, while those of *A. furcatispina* did not. Seeds of the three species showed the same pattern of germination. Germination percentages were significantly lower at 15/5°C than at 25/15 or 35/20°C. The germination temperature pattern found for these species is probably related to the summer seasonal nature of rainfall in the study area. In *A. aroma*, seeds of the two colour morphs showed a similar pattern of dormancy and germination.

Keywords: *Acacia aroma*, *Acacia caven*, *Acacia furcatispina*, germination temperature, physical dormancy

Introduction

Dormancy and germination are very important seed traits that are often considered key elements in plant life history strategies (Rees, 1997). Plants have several

mechanisms to optimize the time of seed germination. These mechanisms favour seedling survival and are very important in natural population dynamics (Bewley and Black, 1994; Rees, 1997).

Seed dormancy caused by a water-impermeable seed coat (i.e. physical dormancy, *sensu* Baskin and Baskin, 1998; Li *et al.*, 1999a, b; Baskin and Baskin, 2004) is common in shrub species that inhabit arid and semi-arid regions (Baskin and Baskin, 1998). Physical dormancy is caused by one or more layers of palisade cells in the seed or fruit coat that are impermeable to water (Baskin *et al.*, 2000). Coat-imposed dormancy can be eliminated by artificial (mechanical or acid scarification) or natural (fire, high temperature, or high fluctuation of temperatures) treatments (Baskin and Baskin, 1998; Jones, 1999; van Assche *et al.*, 2003). In general, when the seed coat becomes permeable to water, seeds can germinate over a wide range of temperatures in both light and darkness (Baskin and Baskin, 1998). In a few species, the embryo also exhibits some degree of physiological dormancy (*sensu* Baskin and Baskin, 1998; Baskin and Baskin, 2004), and seeds can only germinate in a narrow range of temperature or light conditions. Physical plus physiological dormancy is known as combinational dormancy (*sensu* Baskin and Baskin, 1998; Baskin and Baskin, 2004). This type of dormancy has been reported in a small number of species, and its ecological significance remains unclear.

Physical dormancy has been recorded in 15 families of angiosperms, with seven of these including species with physical plus physiological dormancy (Baskin *et al.*, 2000; Baskin, 2003). Physical dormancy is well represented in the three subfamilies of *Fabaceae* (*Caesalpinioideae*, *Mimosoideae* and *Papilionoideae*).

In some species with physical dormancy that inhabit the Mediterranean regions of Australia,

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temperature is crucial for successful germination. For example, several species of *Rhamnaceae* and *Fabaceae* germinate better at low temperatures, coincident with the wet season in this Mediterranean region, than at high temperatures (Bell *et al.*, 1995; Turner *et al.*, 2005). This suggests that germination of these species is related to the time of year that maximizes seedling survival. If this speculation is correct, then in regions with a dry winter and a wet summer, the species should germinate better at higher than at low temperatures.

Although physical dormancy has been reported in several species of *Acacia* (Baskin and Baskin, 1998), the germination requirements of the genus are not clear. *Acacia* (*Fabaceae*, *Mimosoideae*) has a pantropical distribution (Ross, 1981) and includes over 1200 species. In Argentina, *Acacia* is represented by 20 species (Zuloaga and Morrone, 1999) that mainly inhabit arid and semi-arid regions with a monsoon precipitation regime. All the Argentinian species are trees or shrubs and are 2–6 m tall. Our current knowledge about *Acacia* dormancy type and germination requirements is limited. Thus, we studied dormancy and germination in seeds of *Acacia aroma*, *A. caven* and *A. furcatispina*, three important components of the subtropical seasonal xerophytic forest of this country (Cabrera, 1976; Cabido *et al.*, 1994).

Some species of *Fabaceae* show a polymorphism in seed colour. Baskin *et al.* (1998) studied the germination of untreated green and brown seeds of *Senna obtusifolia*. They found higher germination percentages of brown than green seeds, indicating that brown seeds were more permeable than the green ones. They suggested that brown seeds could germinate in the spring in temperate regions, whereas green ones could not germinate until late spring–summer, when temperatures were high enough to cause the seed coat to become permeable. The ecological significance of seed colour polymorphism must be interpreted with caution, and at this moment it is very difficult to establish any pattern (Baskin *et al.*, 1998). *Acacia aroma* produces dimorphic coloured seeds (brown and green).

Here, we present the first of a series of reports in which the dormancy and germination characteristics of seeds from some species of arid and semi-arid regions in Argentina are studied. The aims of this study were to determine whether the seeds of the three *Acacia* species mentioned above have impermeable seed coats, and to describe their germination characteristics. Our hypotheses are that these species have physical dormancy and that the germination percentages of non-dormant seeds are greater at high than at low temperatures. Additionally, we investigated the dormancy and germination characteristics of brown and green seeds of *A. aroma*.

Materials and methods

Species

Acacia aroma Guilles ex Hook. & Arn., *A. caven* (Molina) Molina and *A. furcatispina* Burkart are present in arid and semi-arid regions between 0 and 1500 m above sea level (Zuloaga and Morrone, 1999). In Argentina, they inhabit mainly subtropical seasonal xerophytic forests. The climate in the area is predominantly semi-arid with a monsoon regime (Capitanelli, 1979).

Mature, dry seeds of *A. caven* and *A. aroma* were collected from 20 individuals in the Sierras Chicas, Córdoba province (Argentina), at about 700 m above sea level in March 2004 and May 2004, respectively. Seeds of *A. furcatispina* were collected from 20 individuals in Chancaní, a natural reserve in western Córdoba province (Argentina), in March 2004. The seeds were stored in the laboratory at ambient conditions for 8–15 d before studies were initiated. In seed collections of *A. aroma*, both green (11%) and brown (89%) seeds were present. Considering these proportions, for water imbibition and germination experiments, the number of seeds for the different colour morphs in each replicate was different: 20 for brown and 10 for green seeds. Brown seeds of *A. aroma* were also used for the comparisons between the three species.

Water imbibition

Two treatments – scarified (individually with a razor blade) and non-scarified – were conducted for seeds of each species. Each treatment consisted of three replicates of 20 seeds each for the three species. In the case of green seeds of *A. aroma* (see above), each replicate consisted of 10 seeds. Seeds were placed in 9 cm diameter Petri dishes on filter paper moistened with distilled water. Dishes were placed in a chamber at $22 \pm 1^\circ\text{C}$, with a 12/12 h daily photoperiod, using cool white fluorescent light tubes ($38 \mu\text{mol m}^{-2} \text{s}^{-1}$). After 0, 2, 4 and 24 h, the surface of the seeds was blotted dry, the seeds were weighed with a precision balance (0.1 mg) and returned to the moist filter paper in Petri dishes. The amount of water uptake was determined as the actual increase in seed weight and converted to percentages (see Baskin *et al.*, 2004).

Germination experiment

Scarified and non-scarified seeds of the three species were tested in light (12/12 h daily photoperiod, as above) and in continuous darkness (dishes wrapped with a layer of aluminium foil), with 12/12 h alternating temperature regimes of 15/5, 25/15 and

35/20°C. Three replicates of 20 seeds were used for each treatment. In the scarified treatment, each seed was individually scarified with a razor blade. Seeds were placed in 9 cm diameter Petri dishes on filter paper and moistened (when necessary) with distilled water for 15 d. Germination of seeds incubated in darkness was checked at the end of each experiment. After 15 d, ungerminated seeds at each temperature regime were incubated in light at 22°C for 10 additional days. Protrusion of the radicle was the criterion for germination.

Seed polymorphism

Scarified and non-scarified seeds of the two colour morphs of *A. aroma* were tested in light and in darkness at 15/5 and 35/20°C. Three replicates of 20 (brown) and 10 (green) seeds, respectively (see above), were used for each treatment. In the scarified treatment, each seed was individually scarified with a razor blade. Seeds were placed in 9 cm diameter Petri dishes on filter paper and moistened (when necessary) with distilled water for 15 d.

Data analysis

Final germination percentages were compared with analysis of variance (ANOVA), followed by the LSD Fisher a posteriori test (Sokal and Rohlf, 1995). Prior to the analysis, germination percentages were arcsin transformed to stabilize variances.

Results

Water imbibition

After 24 h at 22°C, the mass of scarified seeds of *A. aroma* (green), *A. aroma* (brown), *A. caven* and *A. furcatispina* had increased 124, 117, 67.5 and 67.6%, respectively, while mass of non-scarified seeds had increased 1.8, 5.2, 3.3, and 5.9%, respectively (Fig. 1). After 5 d at 22°C, mean (\pm SE) germination of scarified seeds was 86.7 ± 0.3 , 98.3 ± 1.6 , 96.6 ± 1.6 and $98.3 \pm 1.6\%$, respectively, while it was only 0, 0, 0 and $63.3 \pm 4.4\%$, respectively, for non-scarified seeds.

Germination experiment

Germination of scarified seeds of the three species and non-scarified seeds of *A. furcatispina* changed with temperature, but not with light treatment (Table 1). No germination of untreated seeds of *A. aroma* and *A. caven* was observed, whereas untreated seeds of *A. furcatispina* germinated at all temperatures (Table 2).

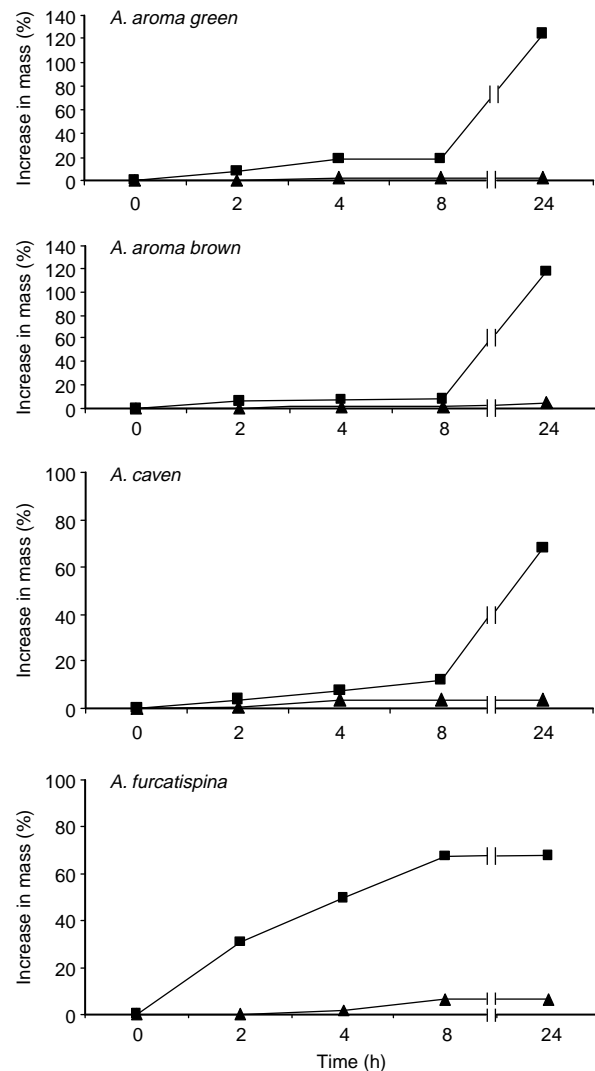


Figure 1. Mean percentage increase in mass of *Acacia aroma* (brown and green seeds), *A. caven* and *A. furcatispina* seeds at 22°C [scarified (■) and non-scarified (▲)].

Scarification promoted germination of *A. aroma* and *A. caven* seeds, and resulted in significantly higher germination of *A. furcatispina* ($P = 0.0021$). However, these significant differences were found only at 15/5°C, both in light and darkness ($P \leq 0.05$, LSD) (Table 2). Germination of seeds of the three species was significantly less at 15/5°C than at 25/15°C and at 35/20°C (Table 2).

Seed polymorphism

Brown and green seeds of *A. aroma* exhibited the same pattern of germination. In both colour types the percentages of germination of scarified seeds were

Table 1. Analysis of variance of the effects for different treatments (temperature regimes and light/dark) on germination of scarified seeds of *Acacia aroma* and *A. caven*, and scarified and non-scarified seeds of *A. furcatispina*

	df	F-value	P
<i>A. aroma</i>			
temperature	2	34.86	0.0001
light	1	0.29	0.61
temperature × light	2	3.27	0.064
<i>A. caven</i>			
temperature	2	62.43	0.0001
light	1	8.44	0.0832
temperature × light	2	2.02	0.1752
<i>A. furcatispina</i> scarified			
temperature	2	12.05	0.0013
light	1	1.06	0.3239
temperature × light	2	1.15	0.3498
<i>A. furcatispina</i> non-scarified			
temperature	2	67.16	0.0001
light	1	2.79	0.121
temperature × light	2	0.74	0.4969

significantly less at 15/5°C compared with 35/20°C, both in light and darkness (Fig. 2). At 15/5°C, green seeds showed a higher percentage of germination than brown ones (Fig. 2). No untreated seeds of either colour morph germinated.

Discussion

Seeds of *A. aroma* and *A. caven* clearly have impermeable coats, as has been reported for many legumes (Bell *et al.*, 1995; Baskin and Baskin, 1998). Non-scarified seeds did not imbibe water, whereas manually scarified seeds took up water in a few hours. These results confirm that these species have physical dormancy imposed by an impermeable seed coat. The hard seed coat may prevent seed germination under unfavourable conditions for seedling development, and it acts as a protection against humidity and temperature fluctuations (Mohamed-Yasseen *et al.*,

1994). In the case of *A. furcatispina*, 63.3% of non-scarified seeds used in the imbibition experiment germinated after 15 d; in this species, this suggested that there are differences in the degree of permeability of the seed coat in seeds of the same lot. Similar results were found in other species of *Fabaceae* (Baskin *et al.*, 1998), *Cistaceae* (Thanos and Georghiou, 1988) and *Malvaceae* (Baskin and Baskin, 1997). The imbibition of scarified seeds occurred at a lower rate in *A. aroma* and *A. caven* than in *A. furcatispina* after 8 h (Fig. 1). These differences could have an ecological significance in terms of species strategy. For example, Turner *et al.* (2005) suggested that the rate of water uptake is related to the habitat in which each species grows; lower water uptake rates are likely to be associated with the prevention of germination at sites where rainfall is sporadic or unpredictable (e.g. drier sites). Even when the three species included in this study coexist in space, *A. furcatispina* is more abundant at the driest site of seed collection, making it difficult to predict the relationships between environmental conditions and water uptake rate in our study area. We believe that this topic calls for further research.

On the other hand, manually scarified seeds of the three study species germinated equally well in light and darkness, but germination was greater at 25/15°C and 35/20°C than at 15/5°C. The pattern of responses of permeable seeds to temperature found in these species may be related to the seasonal nature of rainfall in the study area. The highest amount of rainfall in this region occurs during the summer, when the temperatures are between 13 and 35°C. In the Mediterranean-type climate of Western Australia, where the wet season is the winter, Bell *et al.* (1995) found higher percentages of germination for seeds of *Acacia drummondii* ssp. *candolleana* and *A. lateriticola* at 15°C than at 23°C. Similar results were found by Turner *et al.* (2005) for some Australian *Rhamnaceae*. This suggests that germination of these species appears to be related to the period that maximizes seedling survival. The limited number of seeds germinating at 15/5°C is likely to be related to slow metabolism at low temperatures, rather than the presence of physiological dormancy.

Table 2. Mean percent germination (\pm SE) of scarified (S) and non-scarified (NS) seeds of three *Acacia* species after a 15 d incubation period over a range of daily thermoperiods in light (L) and dark (D). Values with different letters within rows indicate significant differences (LSD, $P \leq 0.05$)

		15/5°C		25/15°C		35/20°C	
		L	D	L	D	L	D
<i>A. aroma</i>	S	3.3 \pm 1.6a	3.3 \pm 1.6a	98.6 \pm 1.6c	88.3 \pm 4.4bc	96.6 \pm 1.6c	98.3 \pm 1.6c
	NS	0	0	0	0	0	0
<i>A. caven</i>	S	10 \pm 7.6a	3.3 \pm 1.6a	98.3 \pm 1.6c	60 \pm 20.2b	98.3 \pm 1.6c	96.6 \pm 1.6c
	NS	0	0	0	0	0	0
<i>A. furcatispina</i>	S	73.3 \pm 14.2ab	50 \pm 7.6a	95 \pm 0bc	85 \pm 10.4bc	96.6 \pm 1.6c	98.3 \pm 1.6c
	NS	43.3 \pm 3.3ab	31.6 \pm 4.4a	75 \pm 5c	60 \pm 0bc	96.6 \pm 3.3d	96.6 \pm 3.3d

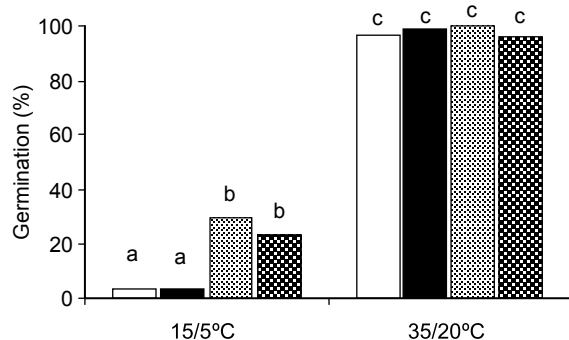


Figure 2. Mean percent germination of brown (solid columns) and green (stippled columns) scarified seeds of *Acacia aroma* after 15 d at 15/5 or 35/20°C in light (white) and dark (black). Different letters indicate significant differences (LSD, $P \leq 0.05$) among mean values of seed germination.

When ungerminated seeds at 15/5°C were transferred to 22°C for 10 d, all of them germinated.

Seeds of the three species germinated equally in light and darkness at all temperature regimes. This result is consistent with previous work showing that seeds with physical dormancy are generally neither inhibited nor promoted by light (Baskin and Baskin, 1998; Baskin *et al.*, 2004).

Brown and green seeds of *A. aroma* exhibited the same pattern of dormancy and germination. These results are not consistent with those reported by Baskin *et al.* (1998) in *Senna obtusifolia*. These authors found that a high percentage of untreated brown seeds were permeable to water. This strategy may allow a developmental window of seed germination for this species. In *A. aroma* the difference in seed colour did not affect its dormancy and germination characteristics. Further work will be necessary to obtain a clear understanding about the dormancy and germination of colour polymorphic seeds.

Physical dormancy is one of the two most common classes of seed dormancy on Earth (Baskin and Baskin, 2003). Seed dormancy and germination requirements are less studied for the arid and semi-arid Argentinian flora. In the present report, we found that the germination requirements of three *Acacia* species were similar, but seed dormancy showed some qualitative differences between them. Seed dormancy was more intense in *A. aroma* and *A. caven* than in *A. furcatispina*. Studying the occurrence of physical seed dormancy in species from different ecosystems will help us to understand its evolution and ecological significance.

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